Quality Timber Products of

Teak

from Sustainable Forest Management

Proceedings of the International Conference on Quality Timber Products of Teak from Sustainable Forest Management
Peechi, India, 2-5 December 2003

Editors

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Published by:

Kerala Forest Research Institute, Peechi 680 653, Kerala, India
Tel: 91-487-2699037, 2699061-64; Fax: 91-487-2699249
Email: kfri@kfri.org Website: http://www.kfri.org
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Email: itto@itto.or.jp Website: http://www.itto.or.jp

ISBN 81-85041-63-6

KFRI Library Cataloguing-in-Publication Data

QUALITY TIMBER PRODUCTS OF TEAK FROM SUSTINABLE FOREST MANAGEMNET

1. Teak products 2. Sustainable forest management-teak
3. Timber certification 4. *Tectona grandis*

I. Bhat, K. M. II Kerala Forest Research Institute
III. International Tropical Timber Organization

582.949

Typeset: KFRI
Printed at Lumiere, Thrissur 680 020, India

Selection and Propagation of Superior Teak for Quality Improvement in Plantations: case study of the ICSB/Cirad-Forêt joint project

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ABSTRACT

Innoprise Corporation Sdn Bhd (ICSB), an investment subsidiary of the Sabah Foundation, has embarked on an extensive Research and Development program for teak (*Tectona grandis*), a highly prized timber species. Through ICSB's joint collaboration with CIRAD-Forêt, a French R and D organization, a plant improvement program was initiated in the early 1990's. Special efforts have been devoted to vegetative propagation strategies based on the development of efficient techniques, both at the laboratory and at the nursery levels. Materials from mature selected plus trees (ortets) from a broad genetic base, and seeds of presumed high genetic value but in restricted number, were multiplied by using a well-developed tissue culture technique at the Plant Biotechnology Laboratory. Additionally, seed lots from natural forest stands, plantations and a multi-provenance clonal seed orchard (CSO) were obtained. These were germinated and used to establish two main provenance/progeny trials in two different locations within ICSB concession in east Sabah, Malaysia, in addition to other numerous conservation and demonstration plots. Data of six-year old trees from these provenance/progeny trials illustrated the great potential of the CSO materials especially in terms of growth rate compared to the other origins under the prevailing conditions. Advanced selection of genotypes combining various traits of major economical value i.e. wood characteristics, is underway. These genotypes and their progenies will be further tested in different sites in the immediate future. The ownership of such a rich germplasm coupled to the efficiency of the developed techniques offers a tremendous potential for genetic improvement, selection and mass production for large-scale plantations of higher yield and superior quality. Commercial production of selected materials from this rich genetic base will then ensue as tissue culture-issued materials for overseas markets.

Keywords: clonal deployment, field trials, genetic resources, germplasm, provenance/progeny trial, superior planting stock production, vegetative propagation, tissue culture.

INTRODUCTION

Teak was first introduced in Kota Marudu in the state of Sabah, Malaysia, by the Dutch Tobacco Company in the 1920s', but did not become popular until the early 1990s (Lapongan, 1998). Since then, private plantation owners and oil palm companies have grown teak mainly by intercropping with oil palm or other cash crops, with particular mention for the mosaic 10x10m interplanted with 10x10m oil palm design experimented at Balung Plantation in Sabah (Salleh, 1995; White, 2002). However, the teak planting material used was not specifically selected for such plantation systems.

Over time, lack of information on the optimal density and spacing in an intercropping system and thereby, fear of potential losses to the yield of associated crops, resulted in the planting of teak mainly along borders of many oil palm plantations and to some extent, roadsides. Moreover, the reputation of long rotation species traditionally associated to teak began to slacken further in Sabah and Malaysia as a whole. As a result, investors nowadays are more eager to invest in the oil palm industry due to the more lucrative returns that can be obtained in a time period as short as three years. Planters were not even encouraged by the finding from a cost analysis of teak planting in Malaysia that estimated teak could be viable as a plantation crop even with an increase of 20% in the cost of planting and a drop in price on a 15-year rotation (Krishnapillay *et al.*, 1997). Nevertheless, while the interest in teak planting within Malaysia declines in the face of competition from other cash crops such as cocoa, rice, rubber and especially oil palm for which the country has for years gained the world production leadership, the scenario is just the opposite in other parts of the world.

Countries in the tropical and sub-tropical zones such as Central and South America, Africa, India, Australia, and other parts of South-east Asia begin embarking on large-scaled planting of teak, recognizing it as a prime candidate for forest plantation establishment in terms of value and world market demand. Accordingly, particular attention is being devoted to means for improving quantitatively and qualitatively the yields in shorter time frames than traditionally expected. Since the early 1990's, Innoprise Corporation, the investment subsidiary of the Sabah Foundation, a state-run organization, and CIRAD-Forêt, a French Research and Development Organization have been working together, aiming at this objective.

CONSTITUTING GENETICALLY-RICH BASE POPULATIONS

Securing the access to a genetically broad-based population is an asset of magnitude importance for developing simple or more sophisticated tree improvement strategies. This is particularly true for teak considering the noticeable differences among provenances, progenies and even half-sib (Bath, 2000). Relevantly, one of our priorities has been to gather as many teak origins as possible and to establish properly designed base population plots within ICSB's concessions.

The first batch of seeds originating from the Solomon island seed-source was introduced in 1989. As for many seed sources, information on the accurate natural origin of this plant material is lacking, although Tenasserim (Myanmar, ex Burma) seems to be the most likely provenance (Kevin White, personal communication). The resulting seedlings were planted by the Plant Improvement and Seed Production (PISP for short) unit as a demonstrational plot in the Luasong Forestry Center, located about two hour drive from Tawau, on the east coast of Sabah. This plant material has been thriving since then, showing remarkable performances under local conditions with average diameter and height of 2-3 cm and 4 m in the first 2-3 years respectively, followed by a gradual evening out in average height of 2 to 3 m in subsequent years. The trees give rise to long clear boles with delayed flowering, bearing in mind that in teak, the later the attainment of the flowering stage, the longer the bole and therefore, the higher the value.

The second introduction of seeds was made in 1995, this time collected from the teak seed stand belonging to the Forest Research Institute of Malaysia in Perlis, located in the north of Peninsular Malaysia close to the border with Thailand. There is still a basic need to document the accurate provenances of these seeds. The small amount of seeds from this batch was germinated under *in vitro* conditions at the Plant Biotechnology Laboratory (PBL) following the procedure described by Monteuuis *et al.* (1998), and then planted as yet another demonstrational plot in Luasong.

In 1996, in order to widen the existing genetic base of teak in our project, ICSB jointly with Cirad-Foret procured seeds from two extensive sources - from natural forest stands or plantations and from progenies produced from a multi-provenance clonal seed orchard in Ivory Coast. Altogether, there were 77 seedlots comprised of India, Thailand, Papua New Guinea, Tanzania, Ivory Coast, Solomon Island, Indonesia, Segama (Sabah) and Perlis (Peninsular Malaysia) (Tables 1 and 2). The seeds were germinated either at the PISP nursery in Luasong or at the PBL, particularly those with presumably low germination capacity. A detailed report on the seedling procedure at the PBL has been published (Monteuuis *et al.*, 1998).

However, in the same way as for many species introduced as exotics, tracing the accurate natural origins of every seed-source as well as the number

Table 1. List of the various seedlots obtained, germinated and planted within our project

Provenances:	* India Chandrapur Maharastra * India Sakrebail Karnataka – 2 batches * India Virnoli Vir. Karnatakan – 2 batches * India Karadibetta Karnataka * India Gilalegundi Karnataka * India Maukal Karnataka – 7 batches * Thailand Mae Huat Lampang (natural stand) * Thailand Mae Huat Lampang (planted stand)
Seed sources:	* Bulu Kumba, Indonesia * Papua New Guinea ex Brown River - presumably from Myanmar (Burma) (Cameron 1966, White, personal communication); * Solomon Island Arara - presumably introduced from Myanmar if not from India or Thailand, via Papua New Guinea (White, personal communication); * Solomon Island Viru–presumably introduced from Myanmar if not from India or Thailand, via Papua New Guinea (White, personal communication); * Perlis, Peninsular Malaysia – from Forest Research Institute Malaysia, presumably from Thailand * Kota Marudu, Sabah – introduced by the Dutch Tobacco Company, presumably from India
	Total: 22 seedlots

of progeny and provenances included, constitute a real problem; information, when reliable, is fragmentary and a lot of uncertainties remain. In this respect, we are placing strong hopes on the use of molecular markers for accessing the information that is lacking.

GERMPLASM CONSERVATION

The teak genetic resources gathered within the project can be conserved either as seeds, planted outdoors or in tissue culture conditions.

As seeds

Seed lots can be stored in a fridge or cold room, at 4°C for some time. However, loss of germination capacity over time must not be underestimated. Several months of storage under such environment may result in a dramatic decrease in germination rate, already known to be low and unpredictable under the best conditions for teak (White, 1991). Depending on the challenge, resorting to tissue culture for germinating recalcitrant seeds can be envisaged, as already successfully undertaken (Monteuuis *et al.*, 1998).

In vivo

Within our project, *in vivo* conservation plots consist mainly in the resources existing in the nursery or in the base and breeding populations, in other words, in demonstration plots, provenance-seed source/progeny trials, clonal tests and seed stands. Practically, mainly genotypes exhibiting superior phenotypes will be preserved. In that sense, our concept of germplasm conservation, deliberately operation-oriented, might be too restrictive. Cost, however, remains for us a major concern. We cannot afford to set up and maintain ex-situ conservation plots for all the genotypes from as many different origins as we can get, owing to various factors such as availability of sites, costs in silvicultural practices and manpower for maintenance of the plots

In vitro

In vitro culture conditions can be a good option for germplasm conservation (Haines, 1994), in a more restricted environment than *in vivo* plots whereby proper maintenance remains a critical issue (Zobel and Talbert, 1984). The pathogen-free characteristics of tissue culture allow the international exchange of living plant

Table 2. List of Families obtained from the Ivory Coast Clonal Seed Orchard and planted within our project

Origin of seed lots			Number of seedlots
India Nellicutha			13
India Nilambur			10
India Vernolirge			3
India Va			2
India Purunakote			3
Ivory Coast Bamoro			2
Ivory Coast Kokondekro			1
Laos Paklay			2
Senegal Djibelor			1
Tanzania Kihuhwi			3
Tanzania Mt			3
Tanzania Bigwa			3
Thailand Huoi-Nam-Oon			3
Thailand Maasale Valley			2
Thailand Pong Salee			2
Thailand Ban Pha Lay			2
Thailand Ban Cham Pui			1
Total :	**17 origins**	**Total :**	**56 seedlots**

material without any sanitary or climatic constraints, contrary to *ex-vitro* plants or plant portions.

Currently, our *in vitro* gene pool is limited to the genotypes under micropropagation. The protocols developed have been conceived as very conservative for maintaining the various genotypes for several years under sustainable sub-culture regimes, while preventing somaclonal variation risks (Monteuuis *et al.*, 1998; Goh and Monteuuis, 2001).

Clonal or genotypic fidelity, which is a requisite for gene bank, remains a crucial concern for us. Another option under investigation currently is the resort to cryopreservation, the efficiency of which has been proven for other tree species (Ashmore, 1997). Developing protocols adapted to teak will be facilitated by the possibility to regenerate teak plant from *in vitro* meristem culture, contrary to many tree species (Monteuuis, unpublished results). The main advantage associated with cryopreservation is the possibility to store genotypes for unlimited periods of times in a very restricted environment and without maintenance requirements, while preserving their integrity, as detailed by Ashmore (1997).

STRATEGIES FOR SETTING-UP IMPROVED YIELD AND QUALITY PLANTATIONS.

From seeds

The traditional means of propagating teak is through seeds, as has been practiced for centuries, with the possibility of storing the seedlings in the form of "stumps" when necessary (Kaosa-ard, 1986; Tin Tun, 2000). Various seed-based strategies have been suggested by specialists for improving the genetic quality of teak seeds (Wellendorf and Kaosa-ard, 1988; Kaosa-ard, 1998, 1999; Kjaer *et al.*, 2000). The seed stand and the seed orchard options are worth considering in our context.

SEED STAND OPTION

The different origins listed (Tables 1 and 2) have been set up on easily accessible ICSB stands according to planting designs adapted to the conversion of the demonstration plots or provenances-seed source/ progeny trial into seed stands. The two provenance/ progeny trials were set up in a partially equilibrated incomplete block design and were comprised of 41 and 42 seedlots respectively, with 26 seedlots common to both trials (Williams and Matheson, 1996). The two sites were very dissimilar in terrain in that one is on a hilly area (Luasong Forestry Center) whereas the other is located on lowland (Taliwas, Lahad Datu). Data from these trials will thereby also allow us to assess the genetic origin x environment interaction. Further, as much as possible, close vicinity of genetically related individuals within each trial has been avoided. Each progeny elementary plot did not include more than 5 siblings in order to minimize the losses when selecting the best in the plot to be kept as seed producers, while felling the others in order to prevent risks of inbreeding depression.

Mass selection prevails at the first step of the selection process. Individual selection is mainly phenotypic, with special attention to traits of major economical importance for teak such as: growth rate, small nodes, bole straightness and basal circularity in absence of buttress or flutes. Wood quality assessment including both aesthetic (wood pattern, texture) and technological characteristics has also to be taken into consideration for phenotypic selection.

In this respect, we are placing a lot of emphasis on the utilization of non-destructive methods for wood quality analysis (Baillères and Durand, 2000).

Further to this selection of plus trees for clonal testing, inferior trees and half-sib in close vicinity are culled in order to favor the intermating of superior individual not genetically related. In the absence of sufficiently detailed records, resort to molecular biology techniques such as AFLP developed in Cirad-Forêt laboratory can help in the determination of the genetic relatedness among individuals within the same neighborhood. The further testing of the selected seed producers of the seed stand based on the performances of their progeny will indicate their combining ability which can be used for refined or more advanced selection and culling activities (Kjaer and Foster, 1996; Mandal and Chawhaan, 1999). A final density of 120 - 180 seed producers per hectare of seed stand is expected at the end of these selection and roguing activities (Figure 1).

SEED ORCHARD OPTION

The best combining genotypes or "combiners" can be asexually propagated or duplicated to be mixed according to a well-suited planting design within a vegetative seed orchard consisting clones from different families and provenances, as illustrated in Figure 1.

These clones will be produced on their own root system as either rooted cuttings, microcuttings or even layering (Lahiri, 1985; Monteuuis *et al.*, 1995; Monteuuis *et al.*, 1998) in order to prevent grafting incompatibility problems and the consequential production of "illegitimates". These are likely to depreciate the genetic quality of the seeds produced, firstly, when collected directly from such unexpected 'mothers' since illegitimates are most of the time hard to distinguish from the grafts as they look similar, and secondly, as these will pollute the genetic quality of the seeds produced by the "legitimates" around.

However, the numerous question marks and uncertainties associated with the real benefits that can be expected from such orchards have to be seriously pondered (Kjaer and Foster, 1996; Kasoa-ard *et al.*, 1998; Kjaer *et al.*, 2000; White and Gavinlertvatana, 1999). The implementation of this strategy is therefore only to be considered for the long term.

IMPROVED QUALITY SEED PRODUCTION STRATEGY

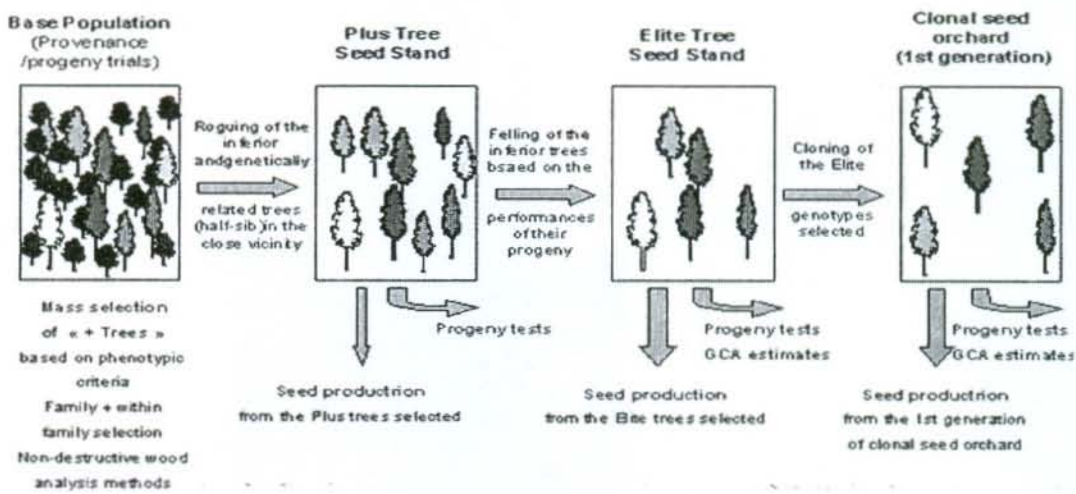

Figure 1: Illustration of the project strategy aiming at producing improved quality seeds in the shortest delay starting from genetically-rich base populations. Advanced generations of clonal seed orchards can be envisaged for the long term.

FROM CUTTINGS OR MICROCUTTINGS

Production of teak planting stock by seeds, although primarily employed, remains severely handicapped by the followings factors:

i. Quantitatively limited seed production (Wellendorf and Kaosa-ard, 1988, White, 1991).

ii. Late flowering. It is noteworthy that in teak, straight bole length, which affects directly its market value, is strictly dependent on the capacity of the terminal meristem to remain vegetative as long as possible (White, 1991). Its conversion into the flowering stage induces a fork formation, as the result of a true dichotomy process.

iii. Low germination rates (Kaosa-ard *et al.*, 1998; White, 1991). In Thailand, for example, only 5 plantable seedlings on average can be practically expected from 100 seeds in a large-scale nursery (Kjaer and Foster, 1996; Kaosa-ard, 1998).

iv. Substantial variability among individuals, even among half-sibs, within progenies, with regard to traits of major economical importance such as growth, form, technological and aesthetic characteristics (wood pattern) (Dupuy and Verhaegen, 1993; Kaosa-ard *et al.*, 1998; Bath, 2000);

v. Limited accurate genetic knowledge about the inheritance of such economically significant traits, and consequently, some uncertainty for the ultimate gain, notwithstanding the time constraints associated with sound breeding programs (Wellendorf and Kaosa-ard, 1988; Kjaer and Foster, 1996; Kaosa-ard, 1998).

The hindrances and uncertainties associated with long term breeding strategies for teak greatly penalized by low seedling productivity have been developed (Kjaer *et al.*, 2000; White and Gavinlertvatana, 1999). According to these authors, the magnitude of the real genetic gain associated with the seedling route has yet to be clearly defined, and the basic question to know whether all the efforts invested during the past decades are worthwhile remains. This is undoubtedly a major concern for potential investors, for which rapid pay-off is a decisive argument. From practical and theoretical information, it can be objectively assumed that for teak, greater genetic gain can be expected from the cutting forestry option, especially when clonal, than that from the seedling forestry (Zobel and Talbert, 1984; Ahuja and Libby, 1993a and b; Monteuuis and Goh, 1999).

Two different strategies can be considered for establishing wood production populations from vegetatively mass-produced teak planting stock, either in bulk form or by clonal propagation.

THE BULK OPTION

Bulk propagation consists in vegetatively propagating a group of mixed genotypes without maintaining any individual identification. This can be useful for increasing the number of a limited quantity of juvenile genotypes of presumably high and similar genetic value, derived for example, from controlled pollination.

The main advantage of the bulk propagation option lies in the absence of a need to strictly and clearly identify by proper labeling one genotype from another. This option will also maintain a certain degree of genetic variability depending on the number of genotypes involved at the beginning, and which in turn may induce an overall heterogeneity in the resulting wood production populations. This seems especially true for teak considering the variability among genotypes. However, successive generations of serial propagation may eventually result in a significant reduction of the original genetic base due to genotypic differences in the multiplication rates, i.e. in the number of shoots produced to be used as cuttings with sufficiently high ability for adventitious rooting.

Bulk propagation has been restricted in our project to superior quality, quantitatively limited or rare seed lots propagated under *in vitro* conditions. At this stage, the genotypes are too young to express any individual difference warranting a selection. The original genetic base in optimized micropropagation conditions, contrary to nursery environment, is thus maintained at least during the first several subculture cycles. However, over time, with little or no control on the genotype-dependent capacity for axillary shoot production, the risk of gradually losing clones with reduced capacity has to be acknowledged.

THE CLONAL OPTION

In clonal propagation, contrary to the bulk propagation strategy, the genotypic identity is rigorously and individually preserved through successive propagation cycles, which may last several centuries in certain cases. Each clone consists of asexually-derived offspring with virtually the same genetic make-up, regardless of the number of its representatives.

Clonal propagation involving scrupulous genotypic identification ensures a better control of the plant material propagated by cuttings than by the bulk option, in addition to a number of other advantages associated with cloning of forest trees (Zobel and Talbert, 1984; Ahuja and Libby, 1993a and b). The possibility to reproduce, theoretically in unlimited numbers, the best trees, from quality and yield standpoints, for large-scale plantation uses, offers tremendous prospects.

The basic requisite for successful teak propagation by rooted cuttings is the existence of a good capacity for adventitious rooting (Monteuuis *et al.*, 1995). From our first field observations, it appears that once rooted, teak cuttings develop "true-to-type", and a good within-clone uniformity can be ultimately expected. Increased yield, higher uniformity for economically important traits such as growth rate, trunk form, straight bole length, wood characteristics, and shorter rotations constitute strong incentives to develop teak clonal plantations (Wellendorf and Kaosa-ard, 1988; Bath, 2000). Such uniformity can not objectively be expected from plantations set up from seedlings, or even from cuttings issued from bulk propagation by virtue of

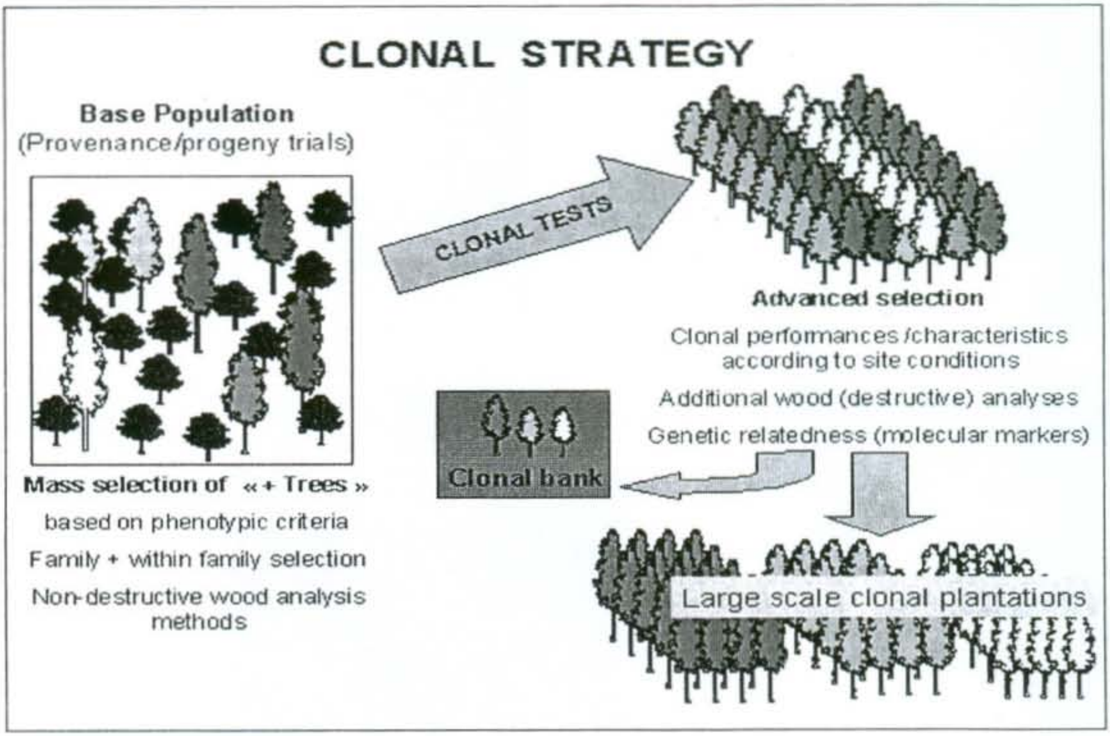

Figure 2: Illustration of the project strategy for large-scale clonal plantations of teak. Molecular markers can help with pedigree information, certification as well as for genetic relatedness between the clones used for clonal deployment.

the arguments developed previously. Properly selected and wisely deployed clones will thus maximize the short-term returns from suitable planting sites, which are dramatically reducing in surface area (Ball *et al.*, 2000).

Utilization of clones can be profitably adapted to intercropping after proper selection particularly, on crown form. Planting density and silviculture practices can be adapted to intensive management systems, with the possibility to harvest several times from the same stump, taking advantage of the excellent coppicing ability (Martin *et al.*, 2000). Such practices look very attractive for enhancing plantation yield while significantly reducing the planting and the (micro)cutting costs. With regard to these financial aspects, it has to be emphasized that clonal plantations generally require less planting stock than plantations established from seedlings, which compensate, to a certain extent, the higher cost of (micro)cuttings. Figure 2 summarizes the clonal strategy adopted for our project.

Efficient techniques for mass vegetative production of superior quality planting stock

Mass production of rooted cuttings in nursery

Mass propagation of teak of any age by rooted cuttings in nursery conditions has been developed and become fully operational within our project since 1992. The requisites, as well as the advantages and limits of this propagation option have been extensively presented and confirmed by a decade of experience (Monteuuis, 1995; Monteuuis *et al.*, 1995; Monteuuis, 2000). Average rooting rates of 80% are routinely obtained from mature genotypes intensively managed as container-grown stock plants once the mobilization phase has been successfully carried out. In such conditions, annual production of 600 rooted cuttings per square meter of stock plant area (15 stock plants per square meter) can be easily obtained (Monteuuis *et al.*, 1995).

In vitro mass propagation

The availability of a well-equipped laboratory within our joint project prompted us to explore the prospects of propagating teak in tissue culture conditions via microcuttings (Bon and Monteuuis, 1996). This successful application has already been reported (Monteuuis *et al.*, 1998; Goh and Monteuuis, 2001). The conceived tissue culture protocols were made as simple as possible in order to be easily applicable and to cope with the constraints of large-scale application in terms of cost efficiency and high productivity. Mass micropropagation of any genotype, either in bulk or clonally, through axillary-produced microshoots with an exponential multiplication rate of three to four at the end of every six week-duration sub-cultures is now possible. The rooting-acclimatization phase is advantageously achieved in nursery conditions under a mist-system with more than 90% success on average. After weaning and raising under the same intensive nursery conditions as for the cutting-derived plants, the tissue-culture plants develop into vigorous and true-to-type vegetative plants. More than 400,000 microcuttings have been produced up to now for local plantations as well as for oversea markets by applying this technique. The possibility to send the *in vitro* plants off to different destinations, regardless of distance, in the absence of phyto-sanitation restrictions, considerably expands the market prospects and constitutes an outstanding asset.

The *in vivo* rooted cutting and *in vitro* microcutting options for vegetatively mass propagating superior teak genotypes with the respective pros and cons have been extensively reviewed, leading to the conclusion that the best option, in many respects, consists in the combination of the two techniques (Monteuuis, 2000).

CONCLUSION AND PROSPECTS

The Research and Development collaborative project between ICSB and Cirad-Forêt was initially implemented to work primarily on rattan species. The shift and the emphasis given to teak started in the early 1990's as a result of the unexpected success obtained from the vegetative propagation techniques developed at the nursery and tissue culture levels. The possibility to mass produce clones by rooted cuttings or *in vitro* microcuttings in cost-effective conditions from any selected mature superior teak tree, as well as from good-performing teak trees introduced locally, prompted us to invest more on these activities. The rationale and the prospects of using clonal materials for teak have been indeed advocated for a long time. Being aware of our vegetative propagation assets, the next step has been to gather our own base populations with a genetic background as rich as possible. This aim has been attained to a large extent by our possession of a highly diverse teak genetic resource that covers the range of adaptability of the species. This noteworthy genetic richness combined to efficient propagation and diffusion techniques are likely to satisfy any plant material order, in the form of seeds, or clones in tissue culture conditions for oversea markets, with the possibility to certify the plant material by DNA fingerprinting.

Keeping up with the latest advances in technology which can further benefit the project remains a major concern. In this respect, we are placing emphasis on the application of molecular markers such as microsatellites and AFLP techniques currently employed for teak by Cirad-Forêt, as well as non-destructive wood analysis methods for moving one step forward in the selection of superior plant material and clonal deployment. Proper site x genotype matching indeed deserves particular attention with respect to pest and disease (defoliators and borers) aspects, as well as to heartwood formation, controlled by genetic and environmental parameters, and which determines log quality (Bailleres and Durand, 2000).

The need to intensify the production of premium quality high value timbers, teak being the most prized one, with a worldwide demand far greater than the supply available, is a strong incentive along this line.

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